NUTRITIONAL CONTENT AND A PHASE - I SAFETY CLINICAL TRIAL OF A HERBAL-NUTRITIONAL SUPPLEMENT (IMUNITI) WITH PUTATIVE IMMUNE-MODULATING PROPERTIES

M.G. Matsabisa[1*], M.P. Sekhoacha[1], O. Ibrahim[2], P. Moodley[3], M. Faber[4]

Email: Motlalepula.matsabisa@mrc.ac.za

[1] IKS Lead Programme, South African Medical Research Council, Francie van Zyl Drive Parow valley, Cape Town, [2] Brenthurst Clinic Parktown Johannesburg, [3] Private GP, Durban South Africa, [4] Nutrition intervention group, South African Medical Research Council Francie van Zijl Drive Parow valley Cape Town

Abstract

The relationship between HIV and AIDS and poor nutrition has been well established. Poor nutrition hastens the progression of HIV infection to AIDS. The rising pandemic of HIV and AIDS and high toxicity associated with anti-retroviral use are major factors that have compelled research to explore traditional herbal medicines as potential alternatives or supplements to anti-retroviral agents. A Phase I clinical trial was conducted on IMUNITI Wellness Pack, a herbal product with putative immune-modulating properties. The product is a combination of 7 herbal preparations, minerals, vitamins, and a specially formulated soya-maize meal porridge and a bottle of water purifier. The aim was to evaluate the safety and tolerability of IMUNITI, with a purpose of developing it for use in HIV-infected patients. The phase I study was conducted at the MRC clinic in Botha's hill and the study lasted 5 weeks from date of participant dosing. The study was a randomised blinded placebo-controlled phase I clinical trial conducted on 48 healthy males. The participants were randomly divided into 4 groups of 12. The 3 groups received different escalating doses of IMUNITI while the forth group received placebo tablets. Participants consumed IMUNITI daily for a period of 5 weeks. Assessments were done at baseline, week 1 and week 5 to determine the safety parameters in all participants. In this study, IMUNITI did not show any safety concerns. In all study participants, there were no significant changes above the upper limit of the reference ranges of the laboratory tests for full blood count, INR, renal and biochemical safety parameters. IMUNITI was well tolerated. Furthermore, the nutritional content analysis of IMUNITI showed that it is a high kilojoule, high protein content product which contains a mixture of sugars, vitamins, traces of calcium, phosphorus and minerals.

Key words: HIV and AIDS, Immune booster, herbal product, traditional medicines

Introduction

An estimated 5.6 million people were living with HIV and AIDS in South Africa in 2009, more than in any other country (UNAIDS 2010). HIV and AIDS accounted for 310,000 deaths in South Africa, resulting in 1.9 million AIDS orphans, of which 330,000 were living with HIV. Prevalence is high among those aged 15-49; almost one-in-three women aged 25-29, and over a quarter of men aged 30-34 are living with HIV (HRSC 2009; Statistics South Africa 2008, 2010).

South Africa has the largest antiretroviral therapy programme in the world, but given the magnitude of the epidemic, access to treatment is low (Meyer-Rath, G *et* al., 2010). Only about 37% of infected people were receiving treatment for HIV. Meanwhile at least 30 people were dying daily due to their inability to access ARVs (Cornell, 2010; Mail and Guardian 2009). Also, starting treatment at a CD4 count of <350 cells/mm^3 according to the WHO guidelines led to increased demand for ARVs (WHO/UNAIDS/UNICEF 2010). The increased deployment of ARVs and an influx of new patients put a strain on the administrative capacity of health authorities, which lead to poor service delivery (Peris, 2010; Health-e 2010). This inadequate supply of ARVs lead to interrupted treatment, which together with the non-adherence to the life-time long therapy, promote risk for drug resistance (Krakovska *et* al., 2007; Harrison, 2009., DoH 2010). These challenges emphasise the need for more affordable, easily accessible short term treatment for HIV and AIDS. One unexplored avenue is traditional medicines and development of medicinal herbal products that have antiviral and immune boosting properties. Strengthening the use of scientifically validated traditional medicines in a comprehensive scientific and clinical-based approach, and provision of good nutrition to HIV and AIDS patients could result in drastic reduction of the burden of HIV and AIDS.

The aim of the study presented here was to evaluate the safety and tolerability of IMUNITI Wellness Pack, with the aim of further developing it for use in HIV-infected patients. IMUNITI Wellness Pack is a nutritional herbal product with putative immune-modulating properties. It is a combination of 7 nutritional and herbal medicinal plants, namely, Sutherlandia, African Potato, Chinese Green Tea, Spirulina, Rooibos supplemented with selenium and Zinc, and with the Aloe vera juice which is supplemented with Vitamin C. In addition to these components, the pack contains a specially formulated soya-maize meal porridge and a bottle of water purifier. There are some various anecdotal evidences of the health benefits of IMUNITI Wellness Pack on HIV infected individuals. This evidence can, however, not be used as a basis for the supply of IMUNITI to the larger public. Credible scientific evidence supporting these anecdotes and the safety of the product under controlled conditions need to be provided if the product is to be made available to the public (De Smet, 1995).

Materials and Methods

Study Participants

The study was conducted at the MRC clinical trial site in Valley of Thousand Hills, Botha's Hill in KwaZulu Natal. Forty eight (48)

http://dx.doi.org/10.4314/ajtcam.v9i3S.3

healthy male volunteers between the ages of 18 to 45 were recruited for the study. Participants were recruited from neighbouring villages in Bothas Hill, Kwa-Zulu Natal, South Africa. All subjects were in good health as determined by medical history, physical examination, vital signs, and clinical laboratory measurements. To be included in the study, the participants had to be able to give informed consent, test sero negative for HIV and be between ≥18 and ≤ 45years of age. The exclusion criteria are summarized as follows: 1) History suggestive of tuberculosis (TB), and Diabetes mellitus. 2) On treatment for chronic illness e.g. diabetes, arthritis, TB etc. 3) On medication for immune modulation, herbal medicines, traditional medicines or multivitamins. 4) Excessive Smoking (nicotine dependence). 5) Excessive alcohol dependence. 6) Sero-positive for HIV. The study was approved by the South African Medical Research Council Ethics Committee. All participants included in the trial gave written prior informed consent to participate. Participants had the right to withdraw at any moment by immediately contacting the investigators to inform them, without obligation to provide reasons.

Study design

This study employed a double-blind placebo-controlled design. The product was tested at three different doses. Participants were randomly assigned to one of four treatment groups of 12 (Table 2).. The first group received a standard recommended daily dose of the supplements (Table 1). The second group and third group each received twice and thrice the recommended daily dose, respectively. The fourth group received the standard IMUNITI Wellness Pack but with placebo tablets containing insipient only. Groups 1 – 3 received 150 ml Aloe Vera juice, but diluted for Groups 1 & 2 (Group 1; 50 ml juice + 100 ml Aloe Vera flavoured water and Group 2; 100 ml juice + 50 ml Aloe Vera flavoured water). Group 4 received 150 ml Aloe Vera flavoured water to blind the treatment. Aloe Vera juice was diluted at the manufacturing stage at Impilo drugs, Kwa-Zulu Natal.

Participants met daily at pre-determined venues to take the study medication under the supervision of the field monitor.The study medication was administered between 7:00 and 10:00 hrs daily for 5 weeks. The porridge included in the IMUNITI Wellness Pack was standard for all four groups. The study blinding was maintained by making the placebo tablets similar in appearance to the active study medication and by using Aloe Vera flavoured water.

Table 1. Recommended daily does of IMUNITI Wellness Pack

Constituent	Strength	Recommended daily	Total Daily intake
Sutherlandia	300mg	2 tablets	600mg
African potato	300mg	1 tablet	300mg
Rooibos incl. Selenium Zinc	175mg 0.2mg 50mg	1 tablet with food	175mg 0.2mg 50mg
Pure Spirulina	500mg	1 tablet	500mg
Chinese Green Tea	150mg	1 tablet	150mg
Aloe vera + Vit C	400mg per 50ml 60mg per 50ml	Take 50ml daily	 60mg
Porridge (Soya + Maize) or	50g 100g	2 heaped tablespoons with water or milk twice daily 4 heaped tablespoons with water or milk	
Blue Gold natural water purifier	Drops		
Tap or bottled water		1 drop per 1 litre and left for 1 minute	
Total daily consumption			1835.2mg

Table 2. Study design and dosage; three study groups received Wellness Pack daily at three different doses while the fourth group received placebo tablets. Porridge was standard for all groups.

Group	Participants (n=48)	Dose/day			
		# Wellness Pack/ day (Active)	Placebo Tablets	Aloe Vera (mL)	Porridge (g)
1	12	ecommended (Recommended as in pack + 12 place tablets	12	50 mL juice + 100 mL water*	100
2	12	x (Recommended Wellness Pack but twice number tablets + 6 placebo tablets)	6	100 mL juice + 50 mL water	100
3	12	x (Recommended Wellness Pack but with three tim the number of tablets)	0	150 mL juice	100
4	12	acebo (Recommended Wellness Pack with 18 place tablets	18	150 mL water	100

Safety Assessments

Assessment of product safety included clinical observation and monitoring of haematological and biochemical parameters; liver function tests as measured by ALT, AST, GGT and LDH, and renal function tests as measured by Urea, Creatinine and electrolytes clearance. Physical examinations, anthropometry, vital sign measurements and blood sampling for clinical laboratory testing were performed at a screening assessment, which occurred within 28 days prior to baseline assessments and administration of the first dose of study medication. Similar follow-up assessments were done at week 1 and week 5. All observed and volunteered adverse events (AEs) were recorded daily by field monitors using health assessment questionnaires and by a clinician on standardized case report forms. An independent monitoring safety committee –

Data and Safety Monitoring Board (DSMB) – was created to evaluate the safety of the product by reviewing AEs encountered. The Division of AIDS Table for grading the severity of Adult Adverse Events (DAIDS grading table) wherein a grading severity scale is provided for each possible laboratory test was utilized for AE reporting in this study. Arrangements were made with hospital management at Grey's Hospital, Pietermaritzburg, for participants needing emergency treatment or hospitalisation.

Data analysis

The effect of each dose at different visits on the clinical, haematology and biochemical variables, were assessed by an Analysis of Variance (ANOVA). Individual comparisons between groups were made using Scheffe's acceptance test for post hoc comparisons. A p-value < 0.05 was indicative of statistical significance. SAS version 9.2 was used for all analysis, including the basic descriptive statistics.

Nutritional content of the IMUNITI Wellness Pack

The nutritional content of IMUNIT wellness pack was evaluated without the porridge. The evaluation was outsourced to the Agricultural Research Council (ARC) analysing laboratory, which holds SANAS accreditation for analyses and an ASM number. The samples received were thoroughly mixed before analysis as per manufacturer's instruction of reconstituting the IMUNITI Pack.

Results

Participants

A total of 90 male participants were available for screening for the study. Participants that tested positive for HIV, had grade 3 laboratory abnormalities or tested positive for hepatitis B were excluded. A total of 48 participants were successfully recruited and only 46 completed the study. Two participants dropped out of the study due to reasons not related to the study drug. There was 95.8% adherence to the study.

Results of laboratory tests

The DAIDS grading table was used as criteria for AE reporting. In general, the volunteers showed values within the limits of normality in the results of the clinical tests in the all visits throughout the study. There were no significant changes in all study participants across all dosage groups in the haemoglobin concentration, the MCV values, iron and ferritin levels. IMUNITI had no effect on the metabolism B12, folate and the absorption of iron from the gastrointestinal tract. INR values were within limits showing that IMUNITI did not inhibit vitamin K absorption in the gastrointestinal tract. None of the biochemical safety parameters were above a grade 2 according to the DAIDS table. Random glucose levels were within normal ranges.

Adverse events

No deaths or serious AEs occurred. No AEs were reported by the field monitors. No study participants reported any significant side-effects such as headaches, nausea, vomiting, diarrhoea, rash, fatigue and sleep disturbances. No AEs or significant changes in physical examination were reported in the Case Report Forms. The overall abnormalities in the biochemical and haematological values using the DAIDS table were considered moderate by study Principle Investigator. There we were no defaulters or discontinuations reported. There was 100% compliance to the study medication and 95.8% adherence to the study.

Nutritional content of the IMUNITI pack

The nutritional content of the IMUNITI Wellness Pack without the porridge is shown in Table 3

Table 3. The nutritional content of IMUNITI. The results relate to RDA of adults above the age of 12. and show the recommended daily dose of the IMUNITI Wellness Pack. Where applicable RDA figures are included for comparison only.

Analysis	Unit	Sample IMUNITI	RDA	
			Male Adults (4 Yrs and olde	aximum Daily consumption of IMUN (1835.2mg)
Calcium		0.22	800 – 1300mg	
Phosphorous		0.29		
Dry matter		94.14		
Moisture		5.86		
Fat (ether extraction)		7.39		
NDFIN		0.045		
Organic matter (Calculated)		89.72		
Starch		35.40		
Water Soluble Carbohydrates		26.54		
Total non- structural carbohydrates		60.40		
Carbohydrates (calculated)		64.20	13g/100g	
Sugars				

Glucose	g/100g	Not detected		
Fructose	g/100g	0.23		0.0042
Sucrose	g/100g	19.28		0.3540
Maltose	g/100g	1.11		0.0204
Lactose	g/100g	Not detected		
Rafinose	g/100g	1.24		0.0228
Stachiose	%	1.51		
Vitamins				
Vit A	mg/100g	0.27	400 – 700mg	0.0037
Vit B1	mg/100g	0.07	0.6 - 1.2mg	0.0013
Vit B2	mg/100g	1.20	0.6 - 1.1mg	0.0022
Vit C	mg/100g	22.25	25 – 100mg	0.0408
Vit E	mg/100g	5.60	7- 15mg te[c]	0.1028
Amino acids				
Arginine	g/100g	1.18		0.0217
Serine	g/100g	0.67		0.0123
Aspartic Acid	g/100g	1.18		0.0217
Glutamic Acid	g/100g	2.74		0.0503
Glycine	g/100g	0.69		0.0127
Threonine	g/100g	0.76		0.0140
Alanine	g/100g	0.62		0.0118
Tyrosine	g/100g	0.72	14 – 24mg/kg	0.0132
Proline	g/100g	1.04		0.0191
HO-Proline	g/100g	0.01		0.0002
Metheonine	g/100g	0.15	13 – 24mg/kg	0.0028
Valine	g/100g	0.85	13 – 28mg/kg	0.0156
Phenylalanine	g/100g	0.82	14 – 28mg/kg	0.0151
Isoleucine	g/100g	0.72	10 – 28mg/kg	0.0132
Leucine	g/100g	1.30	14 – 44mg/kg	0.0239
Histidine	g/100g	1.39	29mg/kg(infants only)	0.2551
Lysine	g/100g	0.97	12 – 49mg/kg	0.0178
Cysteine	g/100g	0.12	13-24mg/kg	0.0022
Tryptophan	g/100g	0.19	3 – 4mg/kg	0.0035
Protein	%	18.13	56g*	
Micro elements				
Aluminium	Mg/100g	1.90		0.0349
Boron	Mg/kg	11.50	UL = 20mg	0.0211
Calcium	Mg/100	159	800 – 1300mg	2.918
Chloride	Mg/100g	878	1..9 – 3.6g	16.11
Chromium	Mg/kg	1.03	15 - 35µg/100g	0.0019
Cobalt	Mg/kg	0.11		0.0002
Flouride	Mg/100g	<15.0	1 – 4mg	<0.2753
Magnesium	Mg/100g	118	130 – 420mg	2.1655
Nitrate	Mg/100g	20.80		3.8172
Nitrite	Mg/100g	<20		<0.3670
Phosphorus	Mg/100g	367	3.8 – 4.7g	6.7352
Potassium	Mg/100g	574	1,2 – 1.5g	10.5340
Sodium	Mg/100g	813	Low 120mg/100g	14.920
Sulphur	Mg/100g	29.70		0.5451
Sulphate	Mg/100g	89		1.633
Antimony	Mg/kg	<0.1		<0.0002
Arsenic	Mg/kg	<0.1		<0.0002
Barium	Mg/kg	144		0.264
Cadmium	Mg/kg	<0.1		<0.0002
Copper	Mg/kg	7.90	440 - 900µg	0.0145
Iron	Mg/kg	171	8 - 18mg	0.3138
Lead	Mg/kg	11.72		0.0022
Manganese	Mg/kg	14.50	2.3mg	0.0266
Mercury	Mg/kg	0.10		0.0002
Molybdenum	Mg/kg	0.93	45µg	0.0017
Nickel	Mg/kg	6.50	UL = 1.0mg	0.0119
Nitrogen	Mg/100g	3.24		0.0597
Selenium	Mg/kg	<0.1	30 - 55µg	<0.0002

http://dx.doi.org/10.4314/ajtcam.v9i3S.3

Strontium	Mg/kg	3		0.0055
Tin	Mg/kg	0.59		0.0011
Titanium	Mg/kg	5.58		0.0102
Vanadium	Mg/kg	<0.3	UL = 1.8mg	<0.0006
Zinc	Mg/kg	170	5 - 12mg	0.3120
Fibre and Energy				
Fibre (crude)	%	1.55	3g/100g	
Neutral detergent fibre	%	4.20		
Dietary fibre (total)	%	5.78	25 – 38g	
Insoluble dietary fibre	%	8.31		
Soluble dietary fibre	%	Not detected		
In vitro digestibility	%	87.73		
ME calculation	MJ/Kg	12.79		0.0235
Energy	MJ/kg	ASM 053	170kJ/100g	0.0326

The Wellness Pack has a high protein content (18%) with a composition of 10 essential amino acids and 9 non-essential amino acids. The product has a low fat content of only 7.4% but a high content of starch (35.40%) and a mixture short chain sugars which contribute to its high kilojoule base (170kJ/100g). Vitamins A, B, C and E, and presence of many micro elements, calcium and phosphorus add to the multi-nutritional value of IMUNITI.

Conclusion

In this study, IMUNITI did not show any safety concerns. There were no clinically significant consistent changes in laboratory parameters in the groups receiving IMUNITI compared to the Placebo group, over the 5 week study period. IMUNITI was well tolerated by all 3 groups. IMUNITI is a high energy and high protein content product with a mixture of essential amino acids, useful Vitamins and minerals which may boost the immune system, and can therefore contribute to the recovery process in AIDS patients. Since the progression of HIV to AIDS especially in developing countries is exacerbated by poor nutrition and unhygienic environment, the product could be recommended as a basis for a sustainable nutritional component for wellness programmes especially in the rural settlements where majority of the population is stricken by poverty and lack of clean water.

The consumption of Imuniti Wellness pack does not pose any danger in exceeding any of the RDA for all the product constituents analysed for.

The heavy metal, pesticides and microbial levels were within acceptable WHO, BP, EMEA, EP and USP standards

No *Staphylococcus. aureus, Pseudomonas aeruginosa*, *Salmonella species* and *Escherichia coli* were detected in the IMUNITI sample analysed and the microbial contamination testing results were all within the WHO specifications for traditional medicinal plants (WHO, 2007).

The safety and nutritional content of studies of IMUNITI have given enough data to support the development of bigger scientific and clinical studies to now investigate its efficacy in patients infected with HIV-1.

References

1. Cornell, M., (2010). Temporal changes in programme outcomes among adult patients initiating antiretroviral therapy across South Africa, 2002-2007. AIDS 24(14): 2263-70

2. De Smet ,P. A., (1995). Health risks of herbal remedies. Drug Safety 13: 81 – 93

3. Harrison, D., (2009). 'An Overview of Health and Health care in South Africa 1994-2010: Priorities, Progress and Prospects for New Gains'. Commissioned by the Henry J. Kaiser Family Foundation Muldersdrift, January 24 - 26 2010. www.samedical.gov.za

4. Health-e (2010). 'Provinces face drug stock outs due to overspending'. www.health-e.org.za/news/article

5. Human Sciences Research Council (2009). 'South African National HIV Prevalence, Incidence, Behaviour and Communication Survey, 2008: A Turning Tide Among Teenagers?' www.hrscpress.ac.za

6. Krakovska, O., Wahl, L.M., (2007). Optimal drug treatment regimens for HIV depend on adherence. Journal of Theoretical Biology. 246 (3): 499-509.

7. Mail & Guardian Online (2009, March). 'Out of stock = out of life'. Mail and Guardian

8. Meyer-Rath, G *et*. al (2010) 'Total cost and potential cost savings of the national antiretroviral treatment (ART) programme in South Africa 2010 to 2017'. XVIII International AIDS conference 2010

9. National Department of Health, South Africa; South Africa National AIDS Council (2010) 'Clinical guidelines: PMTCT (Prevention of Mother-to-Child Transmission)'. www.doh.gov.za/docs/factsheets/guidelines/pmtct.pdf

10. Peris Sean Jones, (2012). Mind the gap: Access to ARV medication, rights and the politics of scale in South Africa. Social Science & Medicine 74(1); 28 - 35

11. Statistics South Africa (2008, October), 'Mortality and causes of death in South Africa, 2006: Findings from death notification' www.statssa.gov.za/Publications/statsdownload.asap?p0319.3&SCH=4254 (Publication date 23/10/2008)

12. Statistics South Africa (2010) 'Mid-year population estimates' www.statssa.gov.za/Publications/P0302/P03022010.pdf

13. UNAIDS (2010) 'UNAIDS report on the global AIDS epidemic' www.unaids.org/downloads/20101123_GlobalReport_em.pdf

14.WHO/UNAIDS/UNICEF (2010) 'Towards Universal Access: Scaling up priority HIV/AIDS interventions in the health sector'. http://www.who.int/hiv/pub/2010progressreport/en/index.html

15. WHO (2007). Guidelines for assessing quality of herbal medicines with reference to contaminants and residues. World Health Organization, Geneva. ISBN 978 924 159444 8

http://dx.doi.org/10.4314/ajtcam.v9i3S.3